

CLAIMS

1. A method for inducing transplantation tolerance including the step of administering a G-CSF derivative, or biologically active fragment, homolog or variant thereof, to a donor cell to be transplanted to a recipient.
- 5 2. The method of claim 1 wherein the G-CSF derivative, or biologically active fragment, homolog or variant thereof, comprises recombinant G-CSF.
3. The method of claim 2 wherein the recombinant G-CSF comprises recombinant human G-CSF.
- 10 4. The method of claim 3 wherein the recombinant human G-CSF comprises recombinant methionyl human G-CSF.
5. The method of claim 4 wherein the recombinant methionyl human G-CSF is non-glycosylated.
6. The method of any one of claims 1 to 5 wherein the G-CSF  
15 derivative, or biologically active fragment, homolog or variant thereof, comprises peg-G-CSF, or biologically active fragment, homolog or variant thereof.
7. The method of claim 6 wherein the G-CSF derivative, or biologically active fragment, homolog or variant thereof, comprises an N-  
20 terminal methionyl residue to which a monomethoxypolyethylene glycol is covalently bound thereto.
8. The method of claim 1 wherein the G-CSF derivative comprises G-CSF or a biologically active G-CSF fragment having a same amino acid sequence as an amino acid sequence of endogenous G-CSF of the donor.

9. The method of any one of claims 1-8 wherein the G-CSF derivative or biologically active fragment, homolog or variant thereof, is administered to the donor cell *in vivo* by administering said G-CSF derivative to a donor.
- 5 10. The method of claim 9 wherein the G-CSF derivative is administered to the donor as a single dose.
11. The method of claim 9 wherein the G-CSF derivative or biologically active fragment, homolog or variant thereof is administered to the donor in a range from 60 µg/Kg weight of the donor-300 µg/kg weight of the
- 10 donor.
12. The method of claim 9 wherein the donor is administered between 6 mg-18 mg of the G-CSF derivative or biologically active fragment, homolog or variant thereof, wherein said donor is human.
13. The method of claim 12 wherein the donor is administered 6
- 15 mg of the G-CSF derivative or biologically active fragment, homolog or variant thereof.
14. The method of claim 9 wherein the donor cell is isolated from the donor after *in vivo* administration of the G-CSF derivative or biologically active fragment, homolog or variant thereof.
- 20 15. The method of any one of claims 1-14 wherein the donor cell comprises a cell obtained from an organ, blood or tissue, a single cell suspension, unseparated cells, enriched cells and homogeneous cells.
16. The method of claim 15 wherein the donor cell comprises an immune cell.

17. The method of claim 16 wherein the immune cell is a T cell.
18. The method of claim 17 wherein administering the G-CSF derivative or biologically active fragment, homolog or variant thereof, stimulates the T cell to produce IL-10.
- 5 19. The method of claim 18 wherein the T cell is MHC class II restricted.
20. The method of claim 16 wherein the immune cell is a granulocyte-monocyte.
21. The method of claim 20 wherein the granulocyte-monocyte is  
10 characterized by a CD11c negative phenotype.
22. The method of claim 21 wherein the granulocyte-monocyte is further characterized by a CD11b<sup>hi</sup>Gr-1<sup>dim</sup> phenotype.
23. The method of claim 22 wherein the donor granulocyte-monocyte is further characterized by a MHC Class I positive, MHC Class II  
15 positive, CD80 positive, CD86 positive and CD40 negative phenotype.
24. The method of claim 23 wherein the granulocyte-monocyte is capable of stimulating a T cell to produce IL-10.
25. The method of claim 24 wherein the T cell is a donor T cell.
26. The method of claim 15 wherein the donor cell comprises a  
20 stem cell.
27. The method of claim 26 wherein the stem cell is obtained from a tissue selected from the group consisting of spleen, blood, bone marrow, skin, nasal tissue and hair follicle.
28. The method of claim 27 wherein the stem cell comprises a

hematopoietic stem cell.

29. The method of any one of claims 14 to 28 wherein the donor cell is isolated and purified as an enriched cell population.

30. The method of claim 29 wherein the enriched donor cell  
5 population comprises a homogeneous cell population.

31. The method of claim 1 wherein the donor cell is isolated from a donor before administering the G-CSF derivative or biologically active fragment, homolog or variant thereof, to the isolated donor cell.

32. The method of any one of claims 29 to 31 further including the  
10 step of propagating the isolated donor cell *in vitro* before transplantation of the donor cell to the recipient.

33. The method of any one of claims 1 to 32 wherein the donor cell is obtained from a mammal.

34. The method of any one of claims 1 to 32 wherein the recipient  
15 is a mammal.

35. The method of claim 33 or claim 34 wherein the mammal is a human.

36. The method of claim 1 wherein transplantation tolerance comprises prevention or reduction of graft versus host disease in the  
20 recipient.

37. The method of claim 36 wherein the prevention or reduction of graft versus host disease is greater than that provided by administering G-CSF to the donor.

38. A method for stimulating a donor T cell to produce IL-10

including the step of administering a G-CSF derivative or biologically active fragment, homolog or variant thereof, to the donor T cell and a donor granulocyte-monocyte to be transplanted to a recipient.

39. The method of claim 38 wherein the G-CSF derivative or  
5 biologically active fragment, homolog or variant thereof comprises recombinant G-CSF.

40. The method of claim 39 wherein the recombinant G-CSF comprises recombinant human G-CSF.

41. The method of claim 40 wherein the recombinant human G-  
10 CSF comprises recombinant methionyl human G-CSF.

42. The method of claim 41 wherein the methionyl human G-CSF is not glycosylated.

43. The method of any one of claims 39 to 42 wherein the G-CSF derivative or biologically active fragment, homolog or variant thereof,  
15 comprises polyethylene glycol.

44. The method of claim 43 wherein the G-CSF derivative, or biologically active fragment, homolog or variant thereof, comprises an N-terminal methionyl residue to which a monomethoxypolyethylene glycol is covalently bound thereto.

20 45. The method of claim 38 wherein the donor granulocyte-monocyte is characterized by a CD11c negative and a CD11b<sup>hi</sup>Gr-1<sup>dim</sup> phenotype.

46. The method of any one of claims 38 to 45 wherein the donor T cell and donor granulocyte-monocyte are obtained from a mammal.

47. The method of claim 46 wherein the recipient is a mammal.
48. The method of claim 46 or claim 47 wherein the mammal is a human.
49. The method of claim 38 wherein the G-CSF derivative or  
5 biologically active fragment, homolog or variant thereof, is administered *in vivo* to a donor before transplantation of the donor T cell to the recipient.
50. The method of claim 38 wherein donor non-immune cells in addition to the donor T cells and donor granulocyte-monocyte are transplanted to the recipient.
- 10 51. The method of claim 50 wherein donor non-immune cells comprise stem cells.
52. A pharmaceutical composition for inducing immunological tolerance when administered to a subject comprising a G-CSF derivative or biologically active fragment, homolog or variant thereof and a  
15 pharmaceutically-acceptable carrier.
53. The pharmaceutical composition of claim 52 wherein the G-CSF derivative or biologically active fragment, homolog or variant thereof comprises recombinant G-CSF.
54. The pharmaceutical composition of claim 53 wherein the  
20 recombinant G-CSF comprises recombinant human G-CSF.
55. The pharmaceutical composition of claim 54 wherein the recombinant human G-CSF comprises recombinant methionyl human G-CSF.
56. The pharmaceutical composition of claim 56 wherein the

recombinant methionyl human G-CSF is not glycosylated.

57. The pharmaceutical composition of any one of claims 52 to 56 wherein the G-CSF derivative comprises peg-G-CSF.

58. The pharmaceutical composition of claim 57 wherein the G-  
5 CSF derivative, or biologically active fragment, homolog or variant thereof, comprises an N-terminal methionyl residue to which a monomethoxypolyethylene glycol is covalently bound thereto.

59. The pharmaceutical composition of any one of claims 52 to 58 wherein immunological tolerance comprises transplantation tolerance and  
10 self-tolerance.

60. The pharmaceutical composition of any one of claims 52 to 58 wherein administering the pharmaceutical composition induces greater immunological tolerance when compared with administering G-CSF.

61. The pharmaceutical composition of any one of claims 52 to 58  
15 wherein said subject is human.

62. A pharmaceutical composition for inducing immunological tolerance in a subject comprising one or more isolated cells having been administered a G-CSF derivative or biologically active fragment, homolog or variant thereof.

20 63. The pharmaceutical composition of claim 61 wherein the isolated cell comprises an immune cell.

64. The pharmaceutical composition of claim 62 wherein the immune cell comprises a T cell.

65. The pharmaceutical composition of claim 64 wherein the T cell

produces IL-10.

66. The pharmaceutical composition of claim 65 wherein the immune cell comprises a granulocyte-monocyte.

67. The pharmaceutical composition claim 66 wherein the  
5 granulocyte-monocyte is characterized by a CD11c negative phenotype.

68. The pharmaceutical composition of claim 67 wherein the granulocyte-monocyte is further characterized by a CD11b<sup>hi</sup>Gr-1<sup>dim</sup> phenotype.

69. The pharmaceutical composition of any one of claims 62-68  
10 wherein said subject is human.

70. The pharmaceutical composition of any one of claims 62-69 wherein immunological tolerance prevents or reduces graft versus host disease.

71. Use of the pharmaceutical composition of any one of claims 52  
15 to 70 to induce immunological tolerance in a patient.

72. A method of transplantation including the steps of:

- 20
- (1) administering to a donor a pharmaceutical composition comprising a G-CSF derivative or biologically active fragment, homolog or variant thereof and a pharmaceutically-acceptable carrier;
  - (2) isolating a cell, tissue or organ from said donor; and
  - (3) transplanting said cell, tissue or organ to a recipient.

73. The method of claim 72 wherein the G-CSF derivative or biologically active fragment, homolog or variant thereof comprise



recombinant G-CSF derivative or biologically active fragment, homolog or variant thereof.

74. The method of claim 73 wherein the recombinant G-CSF derivative or biologically active fragment, homolog or variant thereof  
5 comprise human G-CSF derivative or biologically active fragment, homolog or variant thereof.

75. The method of any one of claims 72-74 wherein the G-CSF derivative or biologically active fragment, homolog or variant thereof comprises peg-G-CSF derivative or biologically active fragment, homolog or  
10 variant thereof.

76. The method of claim 75 wherein the donor and recipient are human.

77. The method of claim 72 including the steps of isolating cells from the donor and propagating the isolated cells in vitro before transplanting  
15 said cells to the recipient.

78. The method of claim 72 wherein transplantation comprises heterologous transplantation whereby the donor and recipient are different individuals.

79. The method of claim 72 wherein transplantation comprises  
20 autologous transplantation whereby the donor and recipient are the same individual.

80. A method for inducing self-tolerance in a patient including the step of administering a G-CSF derivative or biologically active fragment, homolog or variant thereof, to the patient.

81. The method of claim 80 wherein inducing self-tolerance in the patient prevents, treats or reduces an autoimmune disorder of the patient.
82. The method of claim 80 wherein the patient is asymptomatic of an autoimmune disorder.
- 5 83. The method of claim 81 or claim 82 wherein the autoimmune disorder is selected from the group consisting of rheumatoid arthritis, systemic lupus erythematosus, multiple sclerosis and inflammatory bowel disease.
84. The method of claim 80 wherein the G-CSF derivative or  
10 biologically active fragment, homolog or variant thereof stimulates an immune cell of the patient to thereby induce self-tolerance.
85. The method of claim 84 wherein the immune cell comprises a T cell.
86. The method of claim 85 wherein said T cell is stimulated to  
15 produce IL-10.
87. The method of claim 84 wherein the immune cell comprises a granulocyte-monocyte cell.
88. The method of claim 87 wherein said granulocyte-monocyte is characterized by a CD11c negative and CD11b<sup>hi</sup>Gr-1<sup>dim</sup> phenotype
- 20 89. The method of claim 84 wherein the immune cell of the patient is isolated from the patient, propagated *in vitro* and administered to the patient.
90. The method claim 80 wherein the G-CSF derivative or biologically active fragment, homolog or variant thereof comprises peg-G-

CSF or biologically active fragment, homolog or variant thereof.

91. The method of claim 90 wherein the peg-G-CSF comprises peg-human G-CSF or biologically active fragment, homolog or variant thereof.

- 5 92. The method of claim 91 wherein the peg-human G-CSF or biologically active fragment, homolog or variant thereof comprises peg-recombinant human G-CSF or biologically active fragment, homolog or variant thereof.